

IT IS CLAIMED:

1. A method for multigenerational plant trait analysis and associated data management comprising:

- 5 a) generating a random insertion of an insertional mutagen in the genome of a T0 plant, and collecting T1 seed from said T0 plant;
- 10 b) growing T1 plants from the seed collected in (a) under conditions to select transformed T1 plants, and assigning a T1 identification number to each transformed T1 plant selected;
- 15 c) optionally analyzing a transformed T1 plant and recording in an electronic database any mutant traits observed in the transformed T1 plant, wherein a database record of a mutant trait observed in a transformed T1 plant is linked to the T1 identification number assigned to the T1 plant analyzed;
- 20 d) collecting T2 seed from the T1 plant analyzed in (c), and assigning a T2 identification number to said seed, wherein the T2 identification number is linked to the T1 identification number assigned to the T1 plant analyzed in (c);
- 25 e) growing T2 plants from the T2 seed collected in (d); and
- f) analyzing a T2 plant grown in (e) for mutant traits and recording in the database any mutant traits observed in the of the analyzed T2 plant that were not present in its parent T1 plant, wherein a record is generated that associates the information of the analyzed T2 plant to any information recorded about its parent T1 plant.

2. The method of Claim 1 wherein the insertional mutagen is an activation tagging vector.

- 30 3. The method of Claim 2 wherein the activation tagging vector comprises an enhancer selected from the group consisting of a multimerized CaMV 35S enhancer, a figwort mosaic virus enhancer, a peanut chlorotic streak caulimovirus enhancer, and a mirabilis mosaic virus enhancer.

4. The method of Claim 3 wherein the enhancer is a mirabilis mosaic virus enhancer.

5. The method of Claim 1 wherein the T0 plant is selected from the group consisting of *Arabidopsis*, tomato, and rice.

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6. The method of Claim 1 wherein the insertion mutagen encodes a selectable marker selected from the group consisting of an antibiotic resistance gene and an herbicide resistance gene.

10 7. The method of Claim 1 wherein in step (b), prior to assigning T1 identification numbers to transformed plants, transformed plants are transplanted into perimeter wells of a multiwell container comprising a central well in which a barcode is provided, wherein a single perimeter well contains a single T1 plant, and wherein the T1 identification number assigned to each T1 plant in a perimeter well derives from the barcode in the corresponding central well and the relative position of the perimeter well holding said T1 plant.

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8. The method of Claim 7 wherein in step (c) a hand-held electronic data entry device equipped with a barcode scanner is used by an observer to record a mutant trait observed in a T1 plant and scan the barcode in the corresponding central well such that the mutant trait observed and recorded in the hand-held electronic data entry device can be later transferred to the electronic database in association with the T1 identification number of the observed plant.

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9. The method of Claim 1 wherein step (c) includes obtaining a digital image of the transformed T1 plant, entering the digital image into the database, and linking the image entry to the T1 identification number assigned to the imaged T1 plant.

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10. The method of Claim 1 wherein the mutant traits analyzed in (c) and (f) are morphological phenotypes.

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11. The method of Claim 1 wherein the T2 plants are analyzed by performing a directed screen to identify altered resistance to an herbicide.

12. The method of Claim 1 wherein the T2 plants are analyzed by performing a directed screen to identify altered resistance to a pathogen, said pathogen selected from the group consisting of fungus, bacteria, virus, nematode, and insect.

5 13. The method of Claim 1 wherein the T2 plants are analyzed by performing a directed screen to identify altered stress tolerance, said stress selected from the group consisting of drought, salt, and metal.

10 14. The method of Claim 1 wherein the T2 plants are analyzed by performing a directed screen to identify altered level of a biochemical component, said biochemical component selected from the group consisting of vitamins, minerals, amino acids, carbohydrates, lipids, oils, nitrogenous bases, isoprenoids, phenylpropanoids, and alkaloids.

15 15. The method of Claim 1 wherein the mutant traits observed in (c) and (f) are recorded in the electronic database using a predefined vocabulary.

16. The method of Claim 1 wherein the T2 seed collected in (d) is distributed into a plurality of storage containers and stored under conditions that allow long-term recovery of the seeds and generation of T2 plants therefrom.

20 17. The method of Claim 16 wherein each of said storage containers is barcoded to relate the T2 seed contained therein with the corresponding T2 identification number used in the database.

25 18. The method of Claim 1 additionally comprising:

(g) querying the database for a specific mutant trait recorded in step (c) and/or step (f);

30 (h) obtaining T2 seed collected in (d) which is associated with the specific mutant trait queried in (g);

(i) performing a directed screen on the T2 seed obtained in (h) or on plants grown therefrom; and

(j) entering the results of the targeted screen into the database such that the targeted screen results entry is linked to the T2 identification number assigned to the T2 seed.

5 19. The method of Claim 18 wherein the specific mutant trait queried is a morphological trait.

20. The method of Claim 16 wherein steps (a) through (f) are repeated such that essentially every gene in the genome of the plant being analyzed is mutated by an insertional mutagen and a library of seeds that collectively represent saturation of the plant genome with insertional mutagens is generated and contained within the storage containers.

10 21. The method of Claim 20 wherein the T0 plant is *Arabidopsis*.

15 22. The method of Claim 1 wherein a dominant mutant trait is identified by additional steps comprising:

20 (g) collecting pollen from a T2 plant or transformant plant generated therefrom displaying a specific mutant trait;

(h) performing a hybrid cross by pollinating a wild-type plant with the pollen collected in (g);

(i) growing F1 hybrid plants from the hybrid cross in (h); and

25 (j) identifying an F1 plant that displays the specific mutant trait,

wherein the specific mutant trait displayed by the F1 hybrid plant identified in (j) is identified as a dominant mutant trait.

30 23. The method of Claim 1 wherein a candidate gene responsible for a mutant trait is identified by additional steps comprising:

(g) rescuing DNA flanking the insertional mutagen from a T1 or subsequent

generation transformed plant;

(h) identifying at least one candidate gene from the DNA rescued in (g); and

5 (i) identifying a candidate gene identified in (h) that is over-expressed in the transformed plant.

24. The method of Claim 23 wherein the insertional mutagen comprises an enhancer element and the mutant trait is dominant, wherein confirmation that the candidate gene
10 identified in (i) causes the dominant mutant trait is achieved by additional steps comprising:

15 (j) preparing a heterologous gene construct that encodes the candidate gene identified in (i) under control of a heterologous enhancer element capable of effecting mis-expression of said candidate gene;

20 (k) generating a transformed a test plant or explant thereof that is the same species as the T0 plant in (a) with the heterologous gene construct;

25 (l) generating transformed progeny from the transformed test plant or explant thereof generated in (k) that mis-express the candidate gene; and

(m) identifying transformed progeny generated in (l) that display the dominant mutant trait.

24. The method of Claim 24 further comprising:

(n) transforming a test plant or explant thereof that is a different species than the T0 plant in (a) with said heterologous gene construct;

30 (o) generating transformed progeny from the transformed test plant or explant thereof generated in (n) that mis-express the candidate gene; and

(p) identifying transformed progeny generated in (o) that display the dominant mutant phenotype.

26. A system managing plant information for allowing a user to associate a mutant trait
5 and genotype information, comprising:

an electronic database of records representing a library of plants having random genomic insertions of an insertional mutagen, where each record represents one plant and optionally contains information regarding at least one observed mutant trait for that plant, and at least some of the records further contain DNA sequence representing one or more candidate plant genes responsible for one or more of the mutant traits of a corresponding plant; and

10 a graphical user interface comprising

a search engine capable of receiving information regarding at least one user-specified mutant trait or DNA sequence, for searching the database, and

15 a results display area for displaying one or more records identified by the search engine, the displayed information in each record including the identification number of a plant having the specified mutant trait or DNA sequence searched, and at least one of the following: (i) unsearched mutant traits associated with that plant, (ii) a visual representation of the plant, and (iii) one or more DNA sequences representing one or more candidate plant genes responsible for one or more of the searched traits of the plant;

20 whereby the user can associate a searched mutant trait with a plant having other mutant traits or with a plant having candidate modified gene sequences, or can associate searched DNA sequence information with a plant having specific mutant traits.

25 27. The system of claim 26, wherein the mutant trait for a particular plant is selected from the group consisting of an altered morphological trait, altered herbicide resistance, altered pathogen resistance, altered stress resistance, and altered biochemical composition.

30 28. The system of claim 26, wherein the user can specify a mutant trait to the search engine and obtain (i) the identification number of a plant having the searched mutant trait, and (ii) information regarding other mutant traits of that plant.

29. The system of claim 18, wherein the user can specify a DNA sequence to the search engine and obtain (i) the identification number of a plant whose genome contains the searched

DNA sequence, and (ii) information regarding one or more mutant traits of that plant associated with one or more candidate plant genes containing the searched DNA sequence.

30. The system of claim 27, wherein the user further obtains one or more DNA sequences
5 representing one or more candidate plant genes responsible for the searched mutant trait of that plant.

31. A system for facilitating business transactions with a user regarding plant materials or a gene sequence comprising:

10 a database of records representing a library of plants having random genomic insertions of an insertional mutagen, where each record represents one plant and contains information regarding at least one observed mutant-trait for that plant, and at least some of the records further contain DNA sequence information representing one or more candidate plant genes responsible for one or more of the mutant traits of a corresponding plant; and

15 a graphical user interface comprising
a search engine capable of receiving information regarding at least one user-specified mutant trait or DNA sequence, for searching the database,

20 a results display area for displaying one or more records identified by the search engine, the displayed information in each record including the identification of a plant having the specified mutant trait or DNA sequence searched, and further includes: (i) only unsearched mutant traits associated with the plant, (ii) unsearched mutant traits associated with the plant and a candidate gene sequence associated with one or more of the searched mutant traits of the plant, and (iii) unsearched mutant traits associated with the plant and a confirmed gene sequence responsible for one or more of the searched mutant traits of the plant, and

25 an area for displaying a plurality of business arrangements available to a user pertaining to plant material or a plant gene sequence of a plant identified in the database, the business arrangements available to the user being dependent on whether the corresponding record of a particular plant contains information in (i), (ii) or (iii);

30 whereby the user can associate a searched mutant trait with a plant having other mutant traits or with a plant having candidate modified gene sequences, can associate searched DNA sequence information with a plant having specific mutant traits, and can enter into a business arrangement regarding plant material or a particular gene sequence of a plant based on the level of information available for that plant.

32. The system of claim 31, wherein the plurality of business arrangements includes assignment, license and joint venture.

33. A computer-readable medium embodying a program of instructions for execution by a
5 computer for implementing a system for allowing users to associate plant mutant trait and genotype information, the program of instructions comprising instructions for:

10 implementing a database of records representing a library of plants having random genomic insertions of an insertional mutagen, where each record represents one plant and contains information regarding at least one observed mutant-trait for that plant, and at least some of the records further contain DNA sequence information representing one or more candidate plant genes responsible for one or more of the mutant traits of a corresponding plant; and

15 providing a graphical user interface for receiving information regarding at least one user-specified mutant trait or DNA sequence, for searching the database, and

20 displaying one or more records identified in the search, the displayed information in each record including the identification number of a plant having the specified mutant trait or DNA sequence searched, and at least one of the following: (i) unsearched mutant traits of potential interest associated with that plant, (ii) a visual representation of the plant, and (iii) one or more DNA sequences representing one or more candidate plant genes responsible for one or more of the searched mutant traits of the plant;

25 whereby the program of instructions imparts functionality to the executing computer, allowing the user to access and search the database to associate a searched mutant trait with the identification number of a plant having other mutant traits or with the identification number of a plant having candidate modified gene sequences, or to associate searched DNA sequence information with a plant having particular mutant traits of interest, and to view the results of the search.